

UCLA

UCLA Previously Published Works

Title

Eukaryotic origins.

Permalink

<https://escholarship.org/uc/item/1z12j5mn>

Journal

Philosophical transactions of the Royal Society of London. Series B, Biological sciences, 370(1678)

ISSN

0962-8436

Author

Lake, James A

Publication Date

2015-09-01

DOI

10.1098/rstb.2014.0321

Peer reviewed

Review



Cite this article: Lake JA. 2015 Eukaryotic origins. *Phil. Trans. R. Soc. B* **370**: 20140321. <http://dx.doi.org/10.1098/rstb.2014.0321>

Accepted: 5 May 2015

One contribution of 17 to a theme issue
'Eukaryotic origins: progress and challenges'.

Subject Areas:

evolution, genomics, microbiology, taxonomy
and systematics, molecular biology, cellular
biology

Keywords:

eukaryotes, eocytes, evolution, origin,
dawn cell, nucleus

Author for correspondence:

James A. Lake
e-mail: lake@mbi.ucla.edu

Eukaryotic origins

James A. Lake

MCDB Biology and Human Genetics, University of California, 232 Boyer Hall, Los Angeles, CA 90095, USA

The origin of the eukaryotes is a fundamental scientific question that for over 30 years has generated a spirited debate between the competing Archaea (or three domains) tree and the eocyte tree. As eukaryotes ourselves, humans have a personal interest in our origins. Eukaryotes contain their defining organelle, the nucleus, after which they are named. They have a complex evolutionary history, over time acquiring multiple organelles, including mitochondria, chloroplasts, smooth and rough endoplasmic reticula, and other organelles all of which may hint at their origins. It is the evolutionary history of the nucleus and their other organelles that have intrigued molecular evolutionists, myself included, for the past 30 years and which continues to hold our interest as increasingly compelling evidence favours the eocyte tree. As with any orthodoxy, it takes time to embrace new concepts and techniques.

1. From ribosome structures to genes and genomes: the evolution of the eocyte tree

In 1983–1984, Walter Fitch walked into my UCLA office during his sabbatical. His visit changed my scientific life. My laboratory was reconstructing ribosome structures, mapping the locations of their proteins and rRNAs using immunoelectron microscopy, and growing the first three-dimensional crystals of ribosomal subunits [1]. I was intrigued by the unusual ribosomal substructures that we had found in an organism called *Sulfolobus solfataricus* and wanted to understand why the ribosomal substructures found in this prokaryote were very similar to those present in eukaryotes [2].

As I explained my ideas to Walter, he replied in his very direct way that I had it all wrong! But we continued our discussions over many weeks as he taught me how to use parsimony, his favourite method for analysing evolutionary trees. In retrospect, our exciting, collegial arguments gave me a conceptual understanding of evolution that would soon allow us to infer the deep eocyte, i.e. dawn cell, roots of eukaryotes from ribosome structures [3], from gene sequences, and ultimately from genomes.

Our first study analysed three-dimensional ribosomal substructures using parsimony. Because ribosomal substructures evolve much more slowly than gene sequences, we circumvented the long branch attraction (LBA) artefact that can easily confound phylogenetic analyses based upon molecular sequences [4]. That first unrooted eocyte tree (figure 1) based on a single eocyte species, *S. solfataricus*, is still consistent with the rooted trees and rings being derived from gene sequences (figure 2). Currently, four phyla have been discovered/named within the *Eocyta*: the *Aigarchaeota* [7], *Crenarchaeota* [8], *Korarchaeota* [9] and *Thaumarchaeota* [10], as summarized in reference [11].

2. Reconstructing the origin of eukaryotes

At the inception of gene sequencing discovering, the origin of eukaryotes was a major scientific goal. Parsimony and distance approaches were the main methods in use, and very few scientists were aware that these simple methods were vulnerable to error when sequences evolved rapidly.

When parsimony fails, it does so in a recognizable fashion. LBA groups all of the slowly evolving sequences into a 'slow-clade' and all of the rapidly evolving

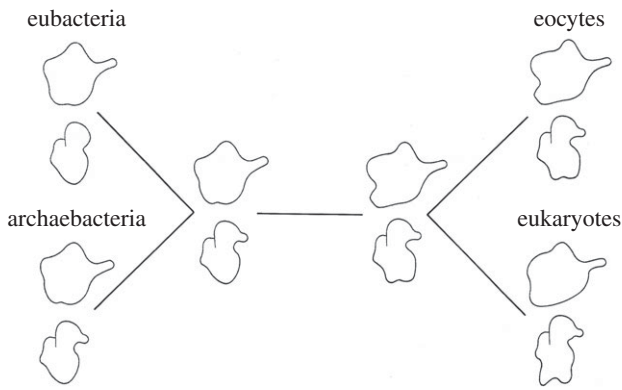


Figure 1. This first ‘eocyte tree’ was reconstructed based on the presence and absence of two ribosomal substructures. These substructures, an additional basal small subunit lobe and an additional lateral large subunit lobe are present exclusively in eukaryotes and in eocytes, and absent in ‘eubacteria’ and ‘archaeobacteria’. Both substructures most parsimoniously support the eocyte tree. Note that this is an unrooted tree. Adapted from [3].

sequences into a ‘fast-clade’. The initial attraction to parsimony when genes were first being sequenced was that it appeared to provide strong, often suspiciously strong, support for LBA trees. In the early days of phylogenetic reconstruction, parsimony’s super strong, but incorrect, results made it the favourite algorithm for studying ‘deep phylogenies’, and even today, LBA-sensitive algorithms are still being used and generating incorrect results.

When eukaryotic 18S ribosomal sequences were first being sequenced, a ferocious competition arose to discover the oldest eukaryote. During this period, every newly sequenced nuclear ribosomal RNA gene was analysed and compared with those from prokaryotes in hopes of discovering the oldest, i.e. the deepest branching, eukaryotic lineage. Every few months, a new sequence analysis would report the discovery of the ‘oldest eukaryote’. The idea that eukaryotes might be very old was exciting and seemed to have strong support.

Ultimately, the race to find the oldest eukaryote collapsed under the weight of the LBA artefact. The final straw leading to the demise of the *Archezoa* occurred when the long-branch leading to the *Microsporidia* (spore-forming intracellular eukaryotic parasites) was nearly an order of magnitude longer than the other branches within the ‘crown group’ of the eukaryotes [12]. Once recognized, this plus the discovery that all *Archezoa* have or once had mitochondria [13] signalled the death knell for the *Archezoa*. But it would take several decades longer for it to be widely accepted that the three domains tree was also caused by LBA.

3. The accumulation of evidence for the eocyte tree over time

I was fortunate to learn about LBA early on because it focused our laboratory on reconstructing evolution in ways that would minimize the effects of LBA. Along the way, we developed several new analytical methods that used novel mathematical approaches to make them less affected by LBA. These included operator metrics, paralinear distances [14], closely related to LogDet which was independently discovered [15], and especially evolutionary parsimony [16] which is based on group theory and should not be confused with parsimony. All of these were more resistant to LBA than

other contemporaneous methods. In 1988, evolutionary parsimony was used to reconstruct the ‘origin of the nucleus’ [17]. That paper was widely covered by the press and it produced letters from around the world, some by anti-evolutionists. I still remember one from a witty fundamentalist who wrote, ‘... you say humans came from an organism that lived at high temperature and smelled of sulfur. I have news for you, that’s not where we came from, that’s where you’re going’.

Although the 1988 paper was quickly challenged [18], a few years later, I was extremely impressed to find that Manolo Gouy, the junior author of the paper that initially challenged eocytes, subsequently published analyses supporting the eocyte tree [19]. This result showed that future leaders in the field were beginning to change their minds as new data were collected and new methods developed. It also gave me hope that the technical details related to LBA were beginning to resonate within the phylogenetic reconstruction community.

In the beginning, the eocyte hypothesis had the support of several leading evolutionary biologists including: Walter Fitch (UC Irvine), Alan Wilson (UC Berkeley) and Colin Patterson (Natural History Museum, London). At that time, few biologists were familiar with the phylogenetic arguments against LBA, so my wife suggested that I apply for funding from the Sloan Foundation to hold winter schools on Evolutionary Biology at UCLA. Similar short courses offered by the MRC in Cambridge had shaped structural biology. I hoped that evolutionary short courses would provide the analytical skills to advance evolutionary biology. These distinguished evolutionists were extremely helpful in getting support from Sloan. The Sloan courses were highly successful and featured speakers such as Wally Gilbert, of DNA sequencing fame, the novelist Irving Stone (who wrote a biography of Darwin), Alan Wilson and Walter Fitch. They helped train a new generation of evolutionary biologists and many of our former students are now leaders in their fields.

Alan Wilson encouraged us to use PCR to sequence ribosomal RNAs and other informational genes from eukaryotes, potential eocytes and reference taxa. Thus, we sequenced many eocyte genes. Among the most useful genes that we sequenced were those coding for protein synthesis elongation factor EF-Tu, because it revealed the existence of an important indel (insertions and deletions within genes) that strongly supported the eocyte evolution of eukaryotes [20]. Even today, those results are so compelling to me that I still do not understand why they were not more widely accepted at the time.

The eocyte controversy also brought with it some unexpectedly positive benefits. It taught us how to quickly sequence genes using PCR, and it also forced us to develop new analytical methods that could handle LBA. Thus, we were positioned to sequence and analyse the relationships between major animal groups using a suite of new tools. The presence of LBA was quickly recognized but we then knew how to circumvent it. As a result we proposed the ‘new animal phylogeny’ that consists of the *Deuterostomia*, the *Lophotrochozoa* and the *Ecdysozoa* [21–23].

Without the eocyte controversy, we might never have discovered the new animal phylogeny, because our success depended upon being able to compensate for LBA. I will never forget the excitement when our evolutionary parsimony calculations first showed that the nematodes and the arthropods were sister taxa—nematodes were then thought to be Aschelminthes. I sat back in my chair almost in shock and

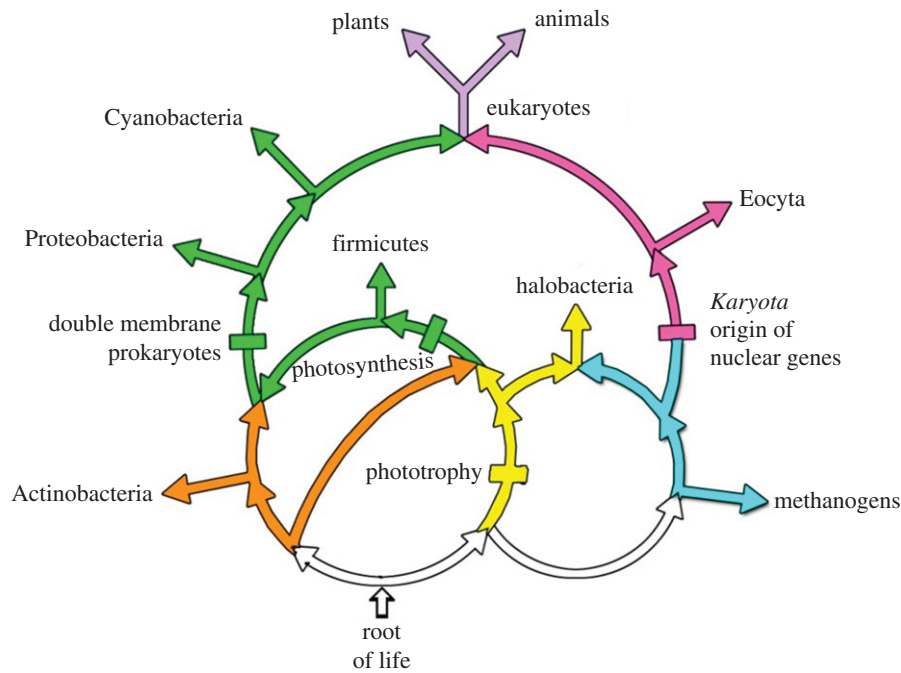


Figure 2. The sister group relationship of the eocytes to the eukaryotes is illustrated by the magenta ‘informational gene flow’ shown on the upper right side of the rings of life. It starts at the rectangle marked ‘Karyota’ and bifurcates to the left to enter the eukaryotes (lavender) and to the right to enter the *Eocyta*. The *Eukaryota* and the *Eocyta* are sister taxa and together form the taxon named the *Karyota*. Formally, the *Eocyta* is the sister taxon to the eukaryotic ‘informational genes’ [5] and the *Karyota* is the clade that includes the *Eukaryota*, the *Eocyta*, and their most recent common ancestor [6].

suddenly realized that these sister taxa both moulted their exoskeletons, and then called my collaborators. For the first time, to the best of my knowledge, a multicellular animal tree had been reconstructed that directly related a genotype to a phenotype (moulting or ecdysis). And it was a huge animal group containing many more species than any other animal super phylum.

In many ways, the new animal phylogeny marked the start of a new phylogenetic era in which LBA was increasingly recognized as a major problem for phylogenetic reconstruction. Because our publications were in highly visible journals, they received much attention, but so did our first eocyte publications. Something was clearly happening in the field of multicellular animal evolution that was different from the first time around. For some reason, the new animal phylogeny met with little resistance, soon entered the textbooks, and was fully accepted by the 150th anniversary of the publication of the *Origin of Species*.

4. The beginnings of an evolutionary renaissance

The eocyte quest also took on some of the aspects of ‘six degrees of separation’. For example, my former graduate student, now Prof. Janet Sinsheimer, and her first graduate student, now Prof. Marc Suchard, developed sophisticated, continuous time Markov models in order to test the eocyte hypothesis [24]. Their methods were precursors to more recent approaches such as NDCH [25] and CAT [26] that by better modelling evolutionary processes led to the recent demonstrations of strong support for the eocyte phylogeny.

In 2008, my wife and I were in Hawaii on holiday when I got an email asking if I would review a manuscript for *PNAS* on the eocyte hypothesis. I was really excited by the abstract, but by the time I got back to the editor, another reviewer had signed on. To me that paper marked the beginning of the resurgence of the

eocyte classification [27]. Since that time, the eocyte hypothesis has been recovered by phylogenetic analyses published by several laboratories using better methods [28–33], so that it has now emerged as the consensus phylogenetic framework for understanding eukaryotic nuclear origins.

5. What are the remaining questions and challenges?

An outstanding challenge is how to relate the eocyte tree and other new findings to eukaryotic evolution more broadly. The eocyte hypothesis deals with the ancestry of the nuclear host lineage and eukaryotic informational genes, but those genes are only one part of the eukaryotic gene complement. Thus, it is clear from our own work [5] and that of others [34–36] that eukaryotic genomes contain many genes for metabolism that are mainly, but not exclusively, of bacterial ancestry. I have argued [6,37] that the chimeric nature of cellular genomes, prokaryotic as well as eukaryotic, can be best understood by a combination of large gene flows and cycle graphs to represent genomic mergers. Our current understanding and hypotheses for the evolution of eukaryotes based upon these ideas and analyses is summarized in figure 2. At least two gene flows merge to form the eukaryotes. These are the informational genes, shown in magenta on the right and the operational genes shown in green on the left [5]. The operational genes are present in eukaryotic chloroplasts and mitochondria, and the informational genes are present in the eukaryotic nucleus. The genes within the informational gene flow underpin the eocyte tree discussed above. The eocytes, formally the *Eocyta* (‘dawn cells’), and the eukaryotes are sister taxa within the eukaryotic informational gene flow as shown in the upper right part of figure 2. Together the eocytes, the eukaryotes and their last common

ancestor form the taxonomic group known as the *Karyota* [38], or the karyotes informally. This sister group based definition of the *Eocyta* provides the phylogenetic basis for experimentally identifying additional eocytes, and suggests clues to the origin and evolution of the nucleus.

The operational gene flow shown in green at the upper left reflects its complex symbiotic origins. The operational gene flow is proposed to have supplied the eukaryotic mitochondria and chloroplasts, and is related to the complex acquisition of these and possibly other eukaryotic cytoplasmic organelles [13]. It is also related to the photosynthetic gene flow [39], shown in green, and to the earlier photrophic gene flow [40] shown in yellow. The rings are rooted at the bottom of figure 2, based on indels incompatible with other possible roots [6].

Other important challenges include studying the early evolution of eukaryotes and more accurately mapping the origins of their informational and operational genes. As we continue to learn more about eukaryotic evolution, we position ourselves to understand the evolution of developmental pathways, in order to relate them to human health, and to understand our evolutionary beginnings. The early evolution of eukaryotes has been complex, and I suspect that the early evolution of humans and other eukaryotes will be equally and possibly far more complex.

Many other major problems are waiting to be solved. Gene divergences and gene convergences of the sort that simultaneously determine both tree-like and ring-like evolution have much to tell us. They can inform us about the deep beginnings of prokaryotes and eukaryotes and they can do it in ways that that can potentially allow us to relate genotypes to phenotypes, but new, improved analytical methods will be needed to reconstruct ring-like evolution.

I am optimistic about the future of evolutionary phylogenomics, especially given the many improvements being made to reduce LBA. I believe that there may be an important story behind each of the gene flows within the rings of life, that those stories may be unlike any that we could have imagined in the past and may simultaneously lead to significant advances in improving human health. I predict that the story will only get better as we understand more about the evolution of life on the Earth. Enjoy the rest of this volume and as you read keep in mind the role of LBA.

As my first departmental chair, George Palade said to me upon his winning the Nobel prize, 'It takes time for new paradigms to displace old orthodoxies, and the decision which is right has to be based on testing, and not on faith'.

Competing interests. We declare we have no competing interests.

Funding. We received no funding for this study.

References

- Lake JA. 1979 *Ribosome structure and functional sites, in ribosomes, structure, function, and genetics*, pp. 207–236. Baltimore, MD: University Park Press.
- Henderson EH, Oakes M, Clark MW, Lake JA, Matheson AT, Zillig W. 1984 A new ribosome structure. *Science* **225**, 510–512. (doi:10.1126/science.6429855)
- Lake JA, Henderson E, Oakes M, Clark MW. 1984 Eocytes: a new ribosome structure indicates a kingdom with a close relationship to eukaryotes. *Proc. Natl Acad. Sci. USA* **81**, 3786–3790. (doi:10.1073/pnas.81.12.3786)
- Felsenstein J. 1978 Cases in which parsimony or compatibility methods will be positively misleading. *Syst. Zool.* **27**, 401–410. (doi:10.2307/2412923)
- Rivera MC, Jain R, Moore JE, Lake JA. 1998 Genomic evidence for two functionally distinct gene classes. *Proc. Natl Acad. Sci. USA* **95**, 6239–6244. (doi:10.1073/pnas.95.11.6239)
- Lake JA, Sinsheimer JS. 2013 The deep roots of the rings of life. *Genome Biol. Evol.* **5**, 2440–2448. (doi:10.1093/gbe/evt194)
- Nunoura T *et al.* 2005 Genetic and functional properties of uncultivated thermophilic crenarchaeotes from a subsurface gold mine as revealed by analysis of genome fragments. *Environ. Microbiol.* **7**, 1967–1984. (doi:10.1111/j.1462-2920.2005.00881.x)
- Woese C, Kandler O, Wheelis M. 1990 Towards a natural system of organisms: proposal for the domains Archaea, Bacteria, and Eucarya. *Proc. Natl Acad. Sci. USA* **87**, 4576–4579. (doi:10.1073/pnas.87.12.4576)
- Elkins JG *et al.* 2008 A korarchaeal genome reveals insights into the evolution of the Archaea. *Proc. Natl Acad. Sci. USA* **105**, 8102–8107. (doi:10.1073/pnas.0801980105)
- Brochier-Armanet C, Boussau B, Gribaldo S, Forterre P. 2008 Mesophilic Crenarchaeota: proposal for a third archaeal phylum, the Thaumarchaeota. *Nat. Rev. Microbiol.* **6**, 245–252. (doi:10.1038/nrmicro1852)
- Guy and Ettema. 2011 The archaeal 'TACK' superphylum and the origin of eukaryotes. *Trends Microbiol.* **19**, 580–587. (doi:10.1016/j.tim.2011.09.002)
- Hirt RP, Logsdon JMJr, Healey B, Dorey MW, Doolittle WF, Embley TM. 1999 Microsporidia are related to fungi: evidence from the largest subunit of RNA polymerase II and other proteins. *Proc. Natl Acad. Sci. USA* **96**, 580–585. (doi:10.1073/pnas.96.2.580)
- Embley TM, Martin W. 2006 Eukaryotic evolution, changes and challenges. *Nature* **440**, 623–630. (doi:10.1038/nature04546)
- Lake JA. 1994 Reconstructing evolutionary trees from DNA and protein sequences: paralinear distances. *Proc. Natl Acad. Sci. USA* **91**, 1455–1459. (doi:10.1073/pnas.91.4.1455)
- Lockhart PJ, Steel MA, Hendy MD, Penny D. 1994 Recovering evolutionary trees under a more realistic model of sequence evolution. *Mol. Biol. Evol.* **11**, 605–612.
- Lake JA. 1987 A rate-independent technique for analysis of nucleic acid sequences: evolutionary parsimony. *Mol. Biol. Evol.* **4**, 167–191.
- Lake JA. 1988 Origin of the eukaryotic nucleus determined by rate-invariant analysis of rRNA sequences. *Nature* **331**, 184–186. (doi:10.1038/331184a0)
- Gouy M, Li WH. 1989 Phylogenetic analysis based on rRNA sequences supports the archaeobacterial rather than the eocyte tree. *Nature* **339**, 145–147. (doi:10.1038/339145a0)
- Tourasse NJ, Gouy M. 1999 Accounting for evolutionary rate variation among sequence sites consistently changes universal phylogenies deduced from rRNA and protein-coding genes. *Mol. Phylogenet. Evol.* **13**, 159–168. (doi:10.1006/mpev.1999.0675)
- Rivera MC, Lake JA. 1992 Evidence that eukaryotes and eocyte prokaryotes are immediate relatives. *Science* **257**, 74–76. (doi:10.1126/science.1621096)
- Lake JA. 1990 Origin of the Metazoa. *Proc. Natl Acad. Sci. USA* **87**, 763–766. (doi:10.1073/pnas.87.2.763)
- Halanych KM, Bacheller JD, Aguinaldo AMA, Liva SM, Hillis DM, Lake JA. 1995 Evidence from 18S ribosomal DNA that the lophophorates are protostome animals. *Science* **267**, 1641–1643. (doi:10.1126/science.7886451)
- Aguinaldo AMA, Turbeville JM, Linford LS, Rivera MC, Garey JR, Raff RA, Lake JA. 1997 Evidence for a clade of nematodes, arthropods and other moulting animals. *Nature* **387**, 489–493. (doi:10.1038/387489a0)
- Suchard MA, Weiss RE, Sinsheimer JS. 2001 Bayesian selection of continuous-time Markov chain

- evolutionary models. *Mol. Biol. Evol.* **18**, 1001–1013. (doi:10.1093/oxfordjournals.molbev.a003872)
25. Foster PG. 2004 Modeling compositional heterogeneity. *Syst. Biol.* **53**, 485–495. (doi:10.1080/10635150490445779)
26. Lartillot N, Philippe H. 2004 A Bayesian mixture model for across-site heterogeneities in the amino-acid replacement process. *Mol. Biol. Evol.* **21**, 1095–1109. (doi:10.1093/molbev/msh112)
27. Cox CJ, Foster PG, Hirt RP, Harris SR, Embley TM. 2008 The archaeobacterial origin of eukaryotes. *Proc. Natl Acad. Sci. USA* **105**, 20 356–20 361. (doi:10.1073/pnas.0810647105)
28. Foster PG, Cox CJ, Embley TM. 2009 The primary divisions of life: a phylogenomic approach employing composition-heterogeneous methods. *Phil. Trans. R. Soc. B* **364**, 2197–2207. (doi:10.1098/rstb.2009.0034)
29. Kelly S, Wickstead B, Gull K. 2011 Archaeal phylogenomics provides evidence in support of a methanogenic origin of the Archaea and a thaumarchaeal origin for the eukaryotes. *Proc. R. Soc. B* **278**, 1009–1018. (doi:10.1098/rspb.2010.1427)
30. Williams TA, Foster PG, Nye TMW, Cox CJ, Embley TM. 2012 A congruent phylogenetic signal places eukaryotes within the Archaea. *Proc. Trans. R. Soc. B* **279**, 4870–4879. (doi:10.1098/rspb.2012.1795)
31. Lasek-Nesselquist E, Gogarten JP. 2013 The effects of model choice and mitigating bias on the ribosomal tree of life. *Mol. Phylogenet. Evol.* **69**, 17–38. (doi:10.1016/j.ympev.2013.05.006)
32. Williams TA, Foster PG, Cox CJ, Embley TM. 2013 An archaeal origin of eukaryotes supports only two primary domains of life. *Nature* **504**, 231–236. (doi:10.1038/nature12779)
33. McInerney JO, O'Connell JO, Pisani D. 2014 The hybrid nature of the Eukaryota and a consilient view of life on Earth. *Nat. Rev. Microbiol.* **12**, 449–455. (doi:10.1038/nrmicro3271)
34. Esser C *et al.* 2004 A genome phylogeny for mitochondria among alpha-proteobacteria and a predominantly eubacterial ancestry of yeast nuclear genes. *Mol. Biol. Evol.* **21**, 1643–1660. (doi:10.1093/molbev/msh160)
35. Cotton JA, McInerney JO. 2010 Eukaryotic genes of archaeobacterial origin are more important than the more numerous eubacterial genes, irrespective of function. *Proc. Natl Acad. Sci. USA* **107**, 17 252–17 255. (doi:10.1073/pnas.1000265107)
36. Martin W, Brinkmann WH, Savona C, Cerff R. 1993 Evidence for a chimeric origin of nuclear genomes: eubacterial origin of eukaryotic glyceraldehyde-3-phosphate dehydrogenase genes. *Proc. Natl Acad. Sci. USA* **90**, 8692–8696. (doi:10.1073/pnas.90.18.8692)
37. Rivera MC, Lake JA. 2004 The ring of life provides evidence for a genome fusion origin of eukaryotes. *Nature* **431**, 152–155. (doi:10.1038/nature02848)
38. Simonson AB, Servin JA, Skophammer RG, Herbold CW, Rivera MC, Lake JA. 2005 Decoding the genomic tree of life. *Proc. Natl Acad. Sci. USA* **102**, 6608–6613. (doi:10.1073/pnas.0501996102)
39. Nelson-Sathi S *et al.* 2012 Acquisition of 1,000 eubacterial genes physiologically transformed a methanogen at the origin of *Haloarchaea*. *Proc. Natl Acad. Sci. USA* **109**, 20 537–20 542. (doi:10.1073/pnas.1209119109)
40. Lake JA, Clark MW, Henderson E, Fay SP, Oakes M, Thornber JP, Mah RA. 1985 Eubacteria, halobacteria, and the origin of photosynthesis: the photocytes. *Proc. Natl Acad. Sci. USA* **82**, 3716–3720. (doi:10.1073/pnas.82.11.3716)